83. (New) The composition of claim 68, wherein the heterologous protein is an antigen of a bacterial cell or a mycobacterial cell.

84. (New) The composition of claim 68, wherein the composition is formulated as a physiologically acceptable composition.

85. (New) The composition of claim 84, further comprising an adjuvant, a pharmaceutically acceptable surfactant, an excipient, a carrier, or a diluent.

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86. 86. (New) The composition of claim 84, wherein the fusion protein is associated with a liposome.

REMARKS

Claim status

The present invention is based, in part, on Applicants' discovery that a discrete domain of a heat shock protein (hsp) can, when joined to a heterologous molecule, produce a $CD8^+$ cytotoxic (cytolytic) lymphocyte (CTL) response in a host to which it is administered.

Claims 36-86 are now pending in the application (claims 1-35 having been canceled by the present amendment and new claims 36-86 having been added). Claim 26, the only independent claim among those previously elected (claims 26-35) covered "[a] composition comprising a heat shock protein, or a portion thereof, joined to a heterologous molecule." The newly added claims cover compositions that include "a portion of" an hsp.

The new claims are supported by the specification. For example, the three new independent claims (**claims 36, 53, and 68**) are supported by the specification at page 3, lines 6-9; pages 9-10; page 11, lines 17-24; original claims 1 and 21; and Figure 12. New **claims 37-38 and 71-72** are supported by the specification at, for example, page 11, lines 17-24, and by original claims 3 and 5. New **claims 39, 54, and 73** are supported by the specification at, for example, page 10, lines 24-29. New **claims 40-43, 55-58, and 74-77** are supported by the specification at, for example, page 10, lines 24-29, Figures 13A, 13B, and 14, and by original

claims 16, 17, and 25. New **claims 44, 59, and 78** are supported by the specification at, for example, page 9, lines 17-20, and by original claim 9. New **claims 45, 60, and 79** are supported by the specification at, for example, page 14, lines 27-30 and page 15, lines 21-25. New **claims 46, 61, and 80** are supported by the specification at, for example, page 8, lines 4-8, and at page 9, lines 17-20. New **claims 47-49, 62-64, and 81-83** are supported by the specification at, for example, page 11, lines 3-9. New **claims 50-52, 65-67, and 84-86** are supported by the specification at, for example, page 12, line 26 through page 13, line 5. New **claims 69-70** are supported by the specification at, for example, page 10, lines 21-23, and by original claim 19. No new matter has been added.

Rejection of Claims 26-35 under 35 U.S.C. § 112, ¶ 2

Claims 26-35 were rejected as being indefinite. More specifically, with respect to claim 26, the Examiner stated, "[i]f applicants wish to claim a particular fragment of a heat shock protein (hsp) please specify the molecule with a defined (*sic.*) sequence structure" (Office action at page 2).

This ground for rejection should be withdrawn in view of the present amendment. Whereas claim 26 (now canceled) referred to "a heat shock protein, or a portion thereof," all of the new independent claims recite "SEQ ID NO:8" – a highly defined sequence. New claims 36 and 53 also cover homologs of SEQ ID NO:8, and new claim 68 includes the term "a substitution mutant or a fragment of SEQ ID NO:8." These terms are also sufficiently definite. One of ordinary skill in the art would recognize homologs or substitution mutants of SEQ ID NO:8 and would certainly be able to discern whether any particular amino acid sequence was a fragment of SEQ ID NO:8.

Claim 26 was also rejected as indefinite because it included the phrase "joined to." The Examiner states that "joined to" "can be explained as mixing two proteins together or connecting two proteins covalently or non-covalently" (Office action at page 2). Applicants respectfully disagree. The usual and accepted meaning of the term "joined," when used in connection with two physical objects (here, two molecules), is that the two objects are physically connected. It is clear that Applicants' claims cover compositions in which a portion of an hsp (*e.g.*, SEQ ID NO:8) is physically connected, by a covalent or non-covalent bond, to a heterologous protein (not

simple mixtures). Moreover, the Examiner is asked to note that new claim 53 specifies that the portion of the hsp and the heterologous protein are fused to one another.

The third reason claim 26 was found indefinite is that it included the term "heterologous molecule." Applicants maintain that the term "heterologous molecule" is clear. Nevertheless, this term has been omitted from all of the presently pending claims. Accordingly, this ground for rejection is moot and should be withdrawn. Similarly, none of the claims now pending include the word "derived," which was also found indefinite, nor do any of the new claims refer to "about half of the ATP binding domain" (Office action at page 3). These grounds for rejection are, therefore, also now moot.

Claim 27 was rejected as unclear because it is the Examiner's opinion that "a portion and an ATP binding domain are not defined" (Office action at page 3). The claims have been amended to clearly indicate that the portion of the hsp is limited to SEQ ID NO:8 or a homolog thereof.

Claim 34 was rejected for inclusion of the term "conservative amino acid substitutions." The Examiner states, "the specification does not teach the definition of a conserved amino acid and fails to disclose which amino acid residue(s) is a conserved amino acid" (Office action at page 3). New claim 69 depends from claim 68 and limits the substitution mutants of claim 68 to those containing "only conservative substitutions of amino acid residues of SEQ ID NO:8". Applicants respectfully request that the prior rejection not be applied to new claim 69. One of ordinary skill in the art would easily recognize a "conservative substitution" of one amino acid for another. Moreover, examples of such substitutions are clearly defined in the present specification. The Examiner's attention is directed to page 10, lines 13-21, where Applicants state:

Amino acid substitutions can be made on the basis of similarity in polarity, charge, solubility, hydrophobicity, hydrophilicity, or the amphipathic nature of the residues involved. For example, the nonpolar (hydrophobic amino acid residues alanine, leucine, isoleucine, valine, proline, phenylalanine, tryptophan, and methionine can be substituted one for another; polar neutral amino acid residues such as glycine, serine, threonine, cysteine, tyrosine, asparagine, and glutamine can be substituted one for another; positively charged (basic) amino acid residues such as arginine, lysine, and histidine can be substituted one for another; and negatively charged

(acidic) amino acid residues such as aspartic acid and glutamic acid can be substituted one for another.

The two final reasons the Examiner found the prior claims indefinite (claim 34 was rejected for including the term "at least" and claim 35 was rejected for the manner in which the term "comprises" was used (Office action at page 3)) are now also moot; the term "at least" does not appear in the present claims (nor does any equivalent term), and the portion of the hsp is well defined as a portion that is limited to SEQ ID NO:8 or a homolog thereof.

In view of the foregoing, Applicants respectfully submit that their claims are clear. One of ordinary skill in the art would be reasonably apprised of their scope. Should the Examiner disagree, Applicants request the favor of a telephone conference to discuss mutually acceptable claim language.

Rejection of Claims 26-35 under 35 U.S.C. § 112 ¶ 1

Enablement. Claims 26-35 were rejected as being inadequately enabled by the specification. The Examiner argues that one of ordinary skill in the art would be forced to conduct undue experimentation in order to practice the full scope of the invention covered by those claims (Office action at pages 3-6). Applicants submit that this ground for rejection should not be applied to the present claims; the specification fully enables one of ordinary skill in the art to make and use the compositions now claimed.

New claims 36 and 68 now recite, as noted above, a defined sequence (SEQ ID NO:8) or a homolog thereof, that is joined (by a covalent or non-covalent bond) to a heterologous protein (not to any molecule, but rather only to a heterologous protein). Notably, SEQ ID NO:8 represents fragment II of *M. tuberculosis* hsp70, which, as the Examiner recognizes, Applicants demonstrated was an effective molecular chaperone. There is no reason why others would have to resort to undue experimentation in order to join SEQ ID NO:8 to a heterologous protein; they could readily make and use such compositions, just as Applicants did. Given the level of skill in the art, one could just as easily select a homolog of SEQ ID NO:8 or modify SEQ ID NO:8 (by deleting one or more of the amino acid residues or replacing them with another residue (see new claim 53). Those of ordinary skill in the art routinely select and modify proteins, and they could readily apply their skills to Applicants' SEQ ID NO:8.

This is not to say that every homolog of SEQ ID NO:8 (or every conceivable fragment or substitution mutant) is now within the scope of the present claims. The compositions now claimed are further limited to those that function as Applicants' fragment II-OVA compositions functioned. That is, to fall within the present claims, a composition must include a homolog of SEQ ID NO:8 (or a fragment or substitution mutant thereof) and a heterologous protein that "when administered to an animal in a physiologically acceptable formulation, elicits a $CD8^+$ cytotoxic T lymphocyte (CTL) response that is greater than the response elicited by administration of the heterologous protein alone". Thus, all of the presently claimed compositions are limited both structurally and functionally, and such compositions are well "enabled" by the specification.

More specifically, with respect to the state of the art and predictability within the art, the Examiner recognizes that hsps can deliver antigens to MHC molecules and induce an antigen-specific CTL immune response (Office action at page 4). The Examiner argues, however, that fusion proteins containing "any and all" fragments of an hsp are unpredictable, and cites, in support of that argument, Geluk *et al.* (*J. Immunol.* 149:2864-2871, 1992; herein, "Geluk"). But Geluk can be legitimately viewed quite differently; what Geluk demonstrates is the high level of skill in the art and the amount of experimentation that is routinely performed. Geluk used a large panel of single amino acid substitution analogs of p3-13 and successfully identified critical HLA-DR17 binding residues. Although Geluk did not make, for example, a substitution mutant of SEQ ID NO:8 (as one might to practice the present invention), her paper demonstrates that molecular biologists routinely make and use great numbers of variant proteins and are well able to assess the importance of particular residues. The amount of experimentation required to practice the present invention is no more than that routinely carried out in this art ("a considerable amount of experimentation is permissible, if it is merely routine" *In re Wands* 858 F.2d 731, 737 (Fed. Cir. 1988) (citing *In re Angstadt*, 537 F.2d 489 (CCPA 1976)). *See also*, the MPEP at 2164.06.

With respect to the scope of the claims and the amount of guidance Applicants provide, we ask the Examiner to consider, as noted above, the new scope of the present claims, which certainly does not extend to compositions containing "any and all fragments of an hsp." The present claims all recite SEQ ID NO:8 (a structural limitation) and all claimed compositions must

elicit a $CD8^+$ cytotoxic T lymphocyte (a functional limitation). Given that Applicants have successfully used a SEQ ID NO:8-containing composition to elicit such an immune response, the scope of the claims is now well aligned with the teaching of the specification. This, considered together with the other "Wands factors," weighs in favor of a finding of enablement. In view of the present scope of the claims, and the remarks provided here, this ground for rejection should now be withdrawn.

Written description. Claims 32-34 were also rejected under 35 U.S.C. § 112, ¶ 1 for lack of an adequate written description. To allow a ready comparison between the claims previously rejected and those now pending, the previously rejected claims (and the claims from which they depended) are reproduced here:

26. (Now canceled) A composition comprising a heat shock protein, or a portion thereof, joined to a heterologous molecule.

27. (Now canceled) The composition of claim 26, wherein the portion of the heat shock protein is a portion of an ATP binding domain of a heat shock protein.

32. (Now canceled) The composition of claim 27, wherein the portion of the ATP binding domain consists of about half of the ATP binding domain.

33. (Now canceled) The composition of claim 26, wherein the portion of the ATP binding domain is a portion of a naturally occurring ATP binding domain in which 1-50% of the amino acid residues have been substituted; 10-40% of the amino acid residues have been substituted; or 10-20% of the amino acid residues have been substituted.

34. (Now canceled) The composition of claim 33, wherein at least half of the substituted amino acid residues are conservative amino acid substitutions.

The Examiner finds that "[t]he specification is rather deficient for teaching when [or] how to make a fusion protein by using a truncated hsp with half [of an] ATP binding domain or

at least 1-50% of substituted conserved amino acids in the ATP binding domain" (Office action at page 6). The Examiner seems to argue that it would not be clear to their colleagues that they were in possession of "any or all fusion protein[s] made by any or all hsp[s] with half [of an] ATP binding (*sic.*) domain or at least 1-50% conserved amino acid residues substituted in the ATP binding domain" (Office action at page 7, lines 9-11). The Examiner then cites passages from *University of California v. Eli Lilly* and *Amgen Inc. v. Chugai Pharmaceutical Co. Ltd.* In view of the present claim amendment, this ground for rejection should be withdrawn. One of ordinary skill in the art, upon reading Applicants' written description, would realize that Applicants fully possessed a particular and effective domain of an hsp (the domain identified as SEQ ID NO:8 – now recited in all of the pending claims), and it would also be evident from the specification that Applicants had adequately described the use of that domain, homologs thereof, or the variants now claimed (*i.e.*, fragments or substitution mutants of SEQ ID NO:8). This would be evident by Applicants' reference to homologs (*e.g.*, at page 10, Applicants refer to hsps from other *Mycobacteria*, as well as to mammalian, fungal, parasitic, and bacterial hsps) or variants of an hsp's ATP binding domain (this would certainly include substitution mutants, which would be clear to one of ordinary skill in the art even without the description of possible variant amino acid residues at page 10 of the written description; see, *e.g.*, the passage reproduced above)).

Contrary to the situation in *U.C. v. Lilly*, the composition Applicants now claim (by structure and function) can be fully visualized. One of ordinary skill in the art would recognize that Applicants were in possession of SEQ ID NO:8, as well as the claimed variants thereof. In view of the present amendment, the Examiner is respectfully asked to reconsider and withdraw the rejection for lack of an adequate written description.

Rejection of Claims 26-31 and 35 under the Judicially Created Doctrine of Double Patenting

Claims 26-31 and 35 were rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1-92 of U.S. Patent No. 6,338,952 in view of Suzue *et al.* (*Proc. Natl. Acad. Sci. USA* 94:13146-13151) (Office action at page 9).

Applicants position is that the claims now pending are patentably distinct from those of U.S. Patent No. 6,338,952. Accordingly, the Examiner is asked to reconsider and withdraw the

rejection for double patenting.

35 U.S.C. § 102

The Examiner has made the following thirteen rejections under paragraphs (b) and (e) of § 102. The rejections are addressed collectively, below. In view of the present claim amendment, Applicants respectfully request reconsideration and withdrawal of all rejections on the basis of anticipation.

The rejections are as follows: **(1)** claims 26, 28-31 and 35 are rejected in view of Young (U.S. Patent No. 6,338,952; Office action at page 10, ¶ 24); **(2)** claims 25-30 are rejected in view of Srivastava (U.S. Patent No. 6,030,618; Office action at pages 10-11, ¶¶ 25-26); **(3)** claims 26 and 28-31 are rejected in view of Rappuoli *et al.* (U.S. Patent No. 6,403,009; Office action at page 11, ¶¶ 27-28); **(4)** claims 26-30 and 35 are rejected in view of Wallen *et al.* (U.S. Patent No. 6,455,493; Office action at page 11, ¶¶ 29-30); **(5)** claims 26-30 are rejected in view of Ciupitu *et al.* (*J. Exp. Med.* 187:685-691, 1998; Office action at pages 11-12; ¶¶ 31-32); **(6)** claims 26-31 and 35 are rejected in view of Young (WO 98/35705; Office action at page 12, ¶¶ 34-35); **(7)** claims 26-31 and 35 are rejected in view of Suzue *et al.* (*Proc. Natl. Acad. Sci. USA* 94:13146-13151, 1997; Office action at page 12, ¶¶ 36-37); **(8)** claims 25-30 are rejected in view of Srivastava *et al.* (WO 95/24923A2; Office action at page 12, ¶¶ 38-39); **(9)** claims 25-31 and 35 were rejected in view of Barrios *et al.* (*Eur. J. Immunol.* 21:2297-2302, 1991; Office action at page 13, ¶¶ 40-41); **(10)** claims 25-31 and 25 are rejected in view of Roman *et al.* (*Immunology* 88:487-492, 1996; Office action at page 13, ¶¶ 42-43); **(11)** claims 26-31 are rejected in view of Ciupitu *et al.* (WO 93/17712A2; Office action at page 13, ¶¶ 44-45); **(12)** claims 26, 28, 29, and 30 are rejected in view of Cohen *et al.* (WO 95/31994A1; Office action at page 13, ¶¶ 46-47); and **(13)** claims 26-35 are rejected in view of Lee *et al.* (WO 98/23735A1; Office action at page 14, ¶¶ 48-49).

In view of the present claim amendment, these grounds for rejection should be withdrawn. None of the publications cited above disclose compositions containing a portion of an hsp that is limited to SEQ ID NO:8 (or to a homolog or other variant thereof (*i.e.*, to a fragment or substitution mutant that functions as required by the present claims). Therefore, none of the publications cited above disclose the compositions now claimed. "A claim is

anticipated only if each and every element as set forth in the claim is found, either expressly or inherently described, in a single prior art reference." *Verdegaal Bros. V. Union Oil Co. of California*, 814 F.2d 628 (Fed. Cir. 1987). *See also* MPEP at 2131.01. Should the Examiner disagree with Applicants' position with respect to this rejection, the favor of a telephone call is requested.

Notice of Change of Correspondence Address

Applicants direct the Examiner's attention to the Notice of Change of Correspondence Address being filed concurrently.

Formal Drawings

Applicants also direct the Examiner's attention to the formal drawings being filed concurrently.

CONCLUSION

In view of the above amendments and remarks, it is believed that all claims are in condition for allowance, and it is respectfully requested that the application be passed to issue. If the Examiner feels that a telephone conference would expedite prosecution of this case, the Examiner is invited to call the undersigned at (978) 341-0036.

Respectfully submitted,

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Dated: June 2, 2003

MARKED UP VERSION OF AMENDMENTS

Specification Amendments Under 37 C.F.R. § 1.121(b)(1)(iii)

Replace the paragraph at page 4, line 23 through page 5, line 2 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure 5A [is] illustrates the P1 peptide amino acid sequence, aligned over a diagram of the hsp65-P1 fusion protein (P1 is shown at the C-terminus of hsp65). When liberated from P1, SIYRYYGL (SEQ ID NO: 1) ([boldface] demarked by arrows) binds to K^b to form the peptide-MHC complex recognized by the 2C TCR. In P1, SIYRYYGL (SEQ ID NO: 1) is flanked 5' and 3' by sequences that lie immediately upstream and downstream, respectively, of peptide bonds that are cleaved (see arrows) in murine cells to liberate naturally occurring peptides (SIINFEKL (SEQ ID NO: 2) from ovalbumin (Ova) and LSPFPFDL (SEQ ID NO: 3) from α-ketoglutaraldehyde dehydrogenase (αKG) (Falk, K., *et al., Eur. J. Immunol., 22*:1323-1326 (1992); Ukada, K., *et al., J. Immunol., 157*:670-678 (1996))).

Replace the paragraph at page 5, lines 5 through12 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure [5C are histograms which provide] 5B is a pair of histograms, which display experimental evidence that P1 and hsp65-P1 are processed intracellularly to yield the SYRGL (SEQ ID NO: 4) octapeptide. 48 hr after transfection with mammalian expression vectors (VR1055 and pCINeo), containing sequences that encode P1 and hsp65-P1, respectively, EL4 cells were incubated for 18 hr with an equal number of naive 2C T cells. Histograms show the percentage of live, $2C^+CD8^+$ cells that were stimulated to upregulate the activation marker CD69. The responses of these naive T cells to control EL4 cells, transfected with the empty (vector) plasmids, are shown as shaded histograms.

Replace the paragraph at page 5, lines 13 through17 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure [5D is a graph showing] 5C is a graph, which displays experimental evidence that normal C57BL/6 mice have T cells that can recognize the SYRGL-K^b complex. A $CD8^+$ T cell line, derived from C57BL/6 mice immunized with the SYRGL (SEQ ID NO: 4) peptide in adjuvant, specifically lysed T2-K^b target cells in a peptide-dependent manner. A highly cytolytic long-term cultured 2C CTL clone (L3.100) is shown for comparison.

Replace the paragraph at page 5, lines 18 through 21 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure 6A is a graph showing [CD8] $CD8^+$ CTL that recognize the SYRGL-K^b complex are produced in C57BL/6 mice injected with hsp65-P1 in PBS but not in those injected similarly with equimolar amounts of various controls (a mixture of P1 and hsp65, the SYRGL (SEQ ID NO: 4) octapeptide, the P1 polypeptide itself, or hsp65 itself; as noted further below, SYRGL is referred to as an "octapeptide" as it is an abbreviation of the sequence SIYRYYGL (SEQ ID NO:1)).

Replace the paragraph at page 5, lines 22 through 26 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure 6B is a graph [showing] illustrating the production of SYRGL-specific CTL in mice injected with [a] various amounts of hsp65-P1, 0.015-1.5 nmoles [(1-100μg)] (1-100 μg) or a control fusion protein in which P1 is linked to the C-terminus of a maltose-binding protein [(Mal-P1, 80μg)] (Mal-P1, 80 μg); lysis of T2-K^b target cells in the absence of added SYRGL (SEQ ID NO: 4) peptide is indicated by unfilled symbols.

Replace the paragraph at page 7, lines 10 through 15 with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figure 9B is a pair of graphs illustrating the behavior of hsp65 fusion protein-activated dendritic cells in vivo. [Activation of dendritic cells in vivo.] Myeloid dendritic cells from lymph nodes draining a subcutaneous site where [hsp654-P1] hsp65-P1was injected 24 hr previously show increased expression of MHC-1 [(Kb)] (K^b) (lower panel) compared to myeloid dendritic cells from lymph nodes draining an uninjected site ("no treatment", upper panel).